IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:		)	Conf. No.: 8669
	Sandrine Bourgeois et al.	)	Art Unit: 1609
Serial No:	10/524,318	)	Examiner:
		)	Aaron J. Kosar
Filed:	February 9, 2005	)	Customer Number:
		)	23448
For:	GALENIC FORM FOR COLONIC DELIVERY OF ACTIVE INGREDIENTS	)	

DECLARATION OF INVENTOR UNDER 37 C.F.R. § 1.132 IN U.S. PATENT APPLICATION NO. 10/524,318

Mail Stop Amendment
Commissioner for Patents
P.O. Box 1450
Alexandria, Virginia 22313-1450

Sir:

I, Antoine Andremont, M.D., Ph.D., declare that:

1. I am an inventor of the subject matter disclosed and claimed in United States Patent Application No. 10/524,318, filed on February 9, 2005, as the national phase entry of PCT (WO 2004/016248), filed February 26, 2004, entitled "Galenic Formulation for Colon Targeted Delivery of Active Principles."

2. I obtained my MD in 1976 at the Medical School of Tours University (Tours, France) and Ph. D. in microbiology in 1986 at the pharmaceutical school of University of Paris 11 (Chatenay-Malabry, France). I am board certified in pediatrics, tropical medicine and microbiology. I have been further trained in epidemiology at the CDC (Atlanta, GA), and have been a research fellow at SUNY (Brooklyn, NY). From 1979 to 1996, I worked

as clinical microbiologist at the Institut Gustave-Roussy, the major French Cancer Research and Treatment Centre. I was appointed Professor of Microbiology at the Pharmacy Medical School of University Paris 11 (Chatenay-Malabry) from 1988 to 1996. Since 1996, I have been a full professor of microbiology, University of Paris 7 Medical School, and head of the bacteriology Laboratory of Bichat-Claude Bernard University hospital, a major referral center in infectious diseases in the Paris area. My research has been carried out successively at the CNRS (UMR8612), the INSERM (U13), and now at EA 6934 University Paris 7.

3. I am author or co-author of more than 130 publications, and I recently published a book in collaboration with the anthropologist Michel Tibon-Cornillot (Le triomphe des bactéries : la fin des antibiotiques? MaxMilo Editions Paris 2007 – translation – "The triumph of bacteria: the end of antibiotics?"). I am listed as an inventor on 4 patents, three of which have generated the creation of a biotech company developing anti-bacterial resistance products currently in testing in animal models. I have been recently participating as expert to the WHO/CIA meeting held in Copenhagen in 2007. I am also currently a consultant with Da Volterra, the assignee of the instant patent application.

4. I am aware that the present Application has been examined by the United States Patent and Trademark Office, that an Office Action was issued on by the United States Patent and Trademark Office on July 2, 2008, and that the claims of the Application as discussed herein have been rejected on various prior art and enablement grounds, where the enablement grounds predominantly relate to the description of the claimed compositions as including "isolated" active agents, rather than whole bacteria which might produce such agents.

5. I have been informed by my legal representatives that the rejections of the claims of the Application can be overcome by presenting evidence to the United States Patent and Trademark Office in support of the enablement of the claims, that the reference cited in the novelty rejection does not include all elements of the claimed

invention, and of the non-obvious differences between using isolated active agents and using bacteria which can produce such active agents, and of the non-obviousness of delivering isolated active agents to the colon.

6. Starting first with the enablement rejections, I note that the enablement rejection is based on the Examiner's apparent concern that the term "isolated" is not defined by the claim, the specification does not provide a "standard for ascertaining the requisite degree" of isolation, and, purportedly, one of skill in the art would not be reasonably apprised of the scope of the invention. I believe there is no factual basis for this rejection.

7. Those of skill in the art well understand the meaning of isolated enzymes, and understand that these are different than the delivery of a biological organism that produces the enzymes. I note that a search on the U.S. Patent and Trademark Office shows thousands of issued patents with the word "isolated" in the claims, which relate to isolated enzymes. Researchers have, for years, known how to isolate enzymes from whole bacteria, and how to use protein expression to produce such enzymes.

8. As I will point out in detail below, the specification clearly teaches a) that there are limitations associated with the prior art colonic delivery of whole bacteria producing β-lactamases (Paragraph 0050); b) examples of isolated enzymes in the literature, and which are commercially available, that can be used to prepare the claimed compositions; c) how to prepare isolated enzymes from whole bacteria; d) how to incorporate the isolated enzymes into pectin beads; e) how to measure enzymatic activity both before and after administration of the drug delivery compositions to ensure that the enzymatic activity was preserved throughout transit; and f) that the compositions described in the specification were capable of achieving the desired drug delivery. One of skill in the art can follow this "roadmap" to produce the claimed compositions.

9. First, the prior art delivery of whole bacteria producing β-lactamases was pointed out in the specification as being unsuccessful. A major limitation associated with

the prior art colonic delivery of whole bacteria that produce β-lactamase enzymes is that they may transfer the drug resistance genes to the commensal flora (See paragraph [0050]). Thus, it is clear that the claimed compositions are intended for delivering isolated enzymes, rather than whole bacteria.

10. Back in 1988, I was listed as a co-inventor in PCT WO/1988/007865. In that patent application, we described using a pharmaceutical composition for oral administration designed to attenuate the effects of β-lactamines on the intestinal flora in humans. The composition contained strict anaerobic bacteria which were non pathogenic in humans and which produced β-lactamases. The composition was intended for co-administration with orally-delivered and systemically-released β-lactam containing antibiotics. Thus, I am an inventor of the prior art colonic delivery of bacteria, I recognized the limitations associated with this approach, and helped develop the approach of delivering the isolated enzymes to the colon.

11. A careful reading of the specification shows that it clearly teaches using isolated enzymes, rather than whole bacteria. For example, each of the working examples that discuss delivering a β-lactamase refer to enzymatic activity (measured in terms of UI/bead). If live bacteria were administered, there would be no mention of enzymatic activity in terms of UI/bead, because the bacteria would simply keep on producing the enzyme (i.e., there would theoretically be unlimited activity, so long as the bacteria remained alive). Thus, the term "UI/bead" only makes sense if one discusses isolated enzymes.

12. The specification also refers to specific isolated enzymes that can be incorporated into the drug delivery compositions. For example, paragraph [0057] specifically lists the erythromycin esterase described in a paper I co-authored, namely, Andremont A. et al.((1985), Infect. Immun. 49 (3), 751), as well as the enzyme capable of inactivating quinolones described by Chen Y et al.((1997) Journal of Industrial Microbiology and Biotechnology 19, 378).

13. The specification further teaches preparing pectin beads encapsulating a commercially-available isolated beta-lactamase. For example, paragraph [0085] states that "[f]or preparation of loaded beads the active ingredient (β-lactamases, penicillinases of type A extracted from Bacillus cereus by **Sigma**) was mixed in with the solution of pectin in a ratio of 3% (Vpa/Vpectin)." (*emphasis added*) As the Examiner is aware, Sigma is one of the largest suppliers of laboratory chemicals and biological agents.

14. The specification further teaches a process for isolating enzymes from whole bacteria. Example 6 shows a process for producing an isolated enzyme. For example, paragraph [0129] teaches that the enzyme erythromycin esterase is an intracellular enzyme, and its solubilization required the cells to be broken (i.e., the bacteria had to be destroyed to obtain the isolated enzyme). This operation was carried out by ultrasonic extraction of centrifuging caps recovered in the potassium phosphate buffer 5 mM, pH 7.5.

15. The working examples (See Examples 1-6) show how to measure the enzymatic activity both before and after delivery. This shows that the enzymes were not only isolated, they were capable of being administered to the colon and still retain enzymatic activity. Oral administration and colonic administration of enzymes, in a way that retains a substantial amount of enzymatic activity, is not a trivial matter. Enzymes are fragile, and lose activity in the gastrointestinal tract if not delivered in a suitable formulation. It is difficult to achieve this end, and also achieve colonic delivery, without having deliver occur (in significant amounts) elsewhere in the digestive tract.

16. Since the specification teaches why one should use isolated enzymes, rather than whole bacteria, where to find isolated enzymes (both commercially and in the literature), how to produce isolated enzymes, and how to measure their activity, one of skill in the art can readily understand what is meant by the term "isolated," as this term is used in the claims.

17. Moving on to the novelty rejection, I note that Claims 12-14, 28-31, 36, 59-62, and 65 were rejected under 35 U.S.C. 102 (b) as anticipated by U.S. Patent No. 6,500,423 to Olshenitsky, as "evidenced by" Arthur, Annales de l'Institut Pasteur, Microbiologie 137(1.1) Jan/Feb 1986, pages 125-134 and Ounissi and Courvalin, P. Gene 1985, 35(3), pages 271-278.

18. First and foremost, Olshenitzky is directed to the delivery of a probiotic (i.e., live bacteria). The active probiotic agent in Olshenitsky is a specific *E. coli* - *E. coli* ATCC Deposit No. 202226. In contrast, the claims are directed to drug delivery compositions which include an isolated active agent capable of inactivating antibiotics. As discussed in great detail above, the use of isolated active agents, rather than whole bacteria, is essential to the invention (again, see Paragraph [0050]). The claimed invention can be distinguished from Olshenitsky in at least two ways. First, the claims require an isolated active agent, such as a beta-lactamase, which can inactivate an antibiotic. This is not the same as a whole bacteria. Second, the Examiner has provided no evidence that this specific *E. coli* even produces a beta-lactamase.

19. The Examiner has cited Arthur, Annales de l'Institut Pasteur, Microbiologie 137(1.1) Jan/Feb 1986, pages 125-134 and Ounissi and Courvalin, P. Gene 1985, 35(3), pages 271-278 for the proposition that probiotics (bacteria) inherently produce erythromycin esterase. This proposition is untrue.

20. I am one of the authors of the Arthur paper cited by the Examiner. In this paper, we constructed a probe specific for the gene ereA of plasmid pIP1100, which confers resistance to erythromycin in those E. coli strains that include this gene. While there are certain E. coli strains that include this gene, this strain is not inherently present in all, or even most, E. coli, let alone the specific E. coli (ATCC Deposit No. 202226) in the cited Olshenitsky reference. Since the premise behind the novelty rejection is incorrect, Ounissi and Courvalin similarly fail to support the Examiner's proposition that the probiotic in Olshenitsky inherently includes an erythromycin esterase. It would appear that a novelty rejection based on a faulty scientific premise should be withdrawn.

21. Moving now to the obviousness rejections, I note that Claims 12-18, 28-36, 45-55, and 59-65 were rejected under 35 U.S.C. 103 (a) as obvious over Sriamoransak, Munjeri, Noguchi, and Ounissi.

22. Sriamoransak and Munjeri are directed to the incorporation of certain active agents into pectinate beads. As stated in the Office Action, neither reference discloses or suggests incorporating agents which inactivate antibiotics into the pectinate beads, nor do they suggest any reason to do so. Noguchi and Ounissi were cited as purportedly teaching isolates of the enzymes erythromycin esterase and macrolide 2'-phosphotransferase I (Mph(A), which are capable of inactivating antibiotics. It appears that the Examiner has concluded that it would be obvious to put any known active agent into any known drug delivery vehicle. I respectfully disagree.

23. While Sriamoransak and Munjeri teach that pectin beads can be used to deliver certain active agents (bovine serum albumin in one case, and an antimalarial compound in the other), they do not teach using pectin beads to deliver active enzymes, let alone enzymes which would inactivate antibiotics. Neither reference provides any reason why one would administer enzymes which inactivate antibiotics specifically to the colon.

24. Noguchi looked at how transcription of the mph(A) gene for macrolide-2'-phosphotransferase I in E. coli is regulated. Noguchi did not disclose or suggest any pharmaceutically beneficial effects of the macrolide-2'-phosphotransferase I enzyme produced by the bacteria, only how its production is regulated.

25. Noguchi's conclusion was that the production of the Mrx and MphR(A) proteins in those E. coli that had an mphA operon should be enhanced in the presence of erythromycin (see the last paragraph on page 5057). This has nothing to do with the colonic delivery of an enzyme that inactivates erythromycin. To the contrary, Noguchi suggests that the presence of erythromycin would enhance the natural production of this

enzyme in those bacterial strains with an mphA operon. Thus, in a patient infected with such a bacteria, the antibiotic would inherently be inactivated in the patient's colon. There would be no need to administer an active agent capable of inactivating the antibiotic to a patient infected with such a bacteria. Further, one of the reasons to administer the active agent to the patient is to prevent the development of a drug resistant strain. If the strain is already there, then the composition's effectiveness is reduced. Thus, if one reads Noguchi as teaching that all patients inherently have such a bacteria in their commensal flora (as the Examiner has apparently done, although such interpretation is factually incorrect), it would lead one away from the claimed invention.

26. Ounissi discloses having cloned and sequenced the ereA gene in certain E. coli strains, which gene is responsible for producing an erythromycin esterase. As with Noguchi, Ounissi does not disclose or suggest any pharmaceutically beneficial effects of the erythromycin esterase enzyme produced by the bacteria, only the sequence for the gene responsible for its production.

27. Neither Noguchi nor Ounissi teach any reason to isolate the enzymes and use them as active pharmaceutical agents, but rather, merely disclose the manner in which the bacteria produce the enzymes. The conventional way that bacteriologists such as myself use information related to how bacteria produce enzymes which inactive antibacterial agents is a) to develop agents which stop the bacteria from producing these enzymes (i.e., so the antibacterial agents will not be inactivated), or b) to produce antibacterial agents which are immune to these enzymes ((i.e., so the antibacterial agents will not be inactivated.

28. The Office Action has not cited any factual or logical basis for its apparent assertion that a bacteriologist would use information on how bacteria inactivate antibiotics to a) develop these agents as active pharmaceutical agents, b) incorporate them into a very specific type of drug delivery vehicle, and c) deliver them specifically to the colon, without any teaching or suggestion as to why one would do this. No such factual or logical basis exists in any of the cited references.

29. There are many types of drug delivery vehicles, which deliver active agents to any desirable location in the body. Compounds administered orally can be specifically delivered to the stomach, the small intestine, or the colon. Compounds can be administered by intravenous, subcutaneous or intramuscular injection, orally, intranasally, sub-lingually, buccally, and the like. The appropriate selection of drug delivery vehicle is determined based on the specific purpose for which the drug is administered and the characteristics of the drug to be incorporated. As a physician, I would never presume that I could administer any active agent in any drug delivery vehicle and have any expectation of success. Rather, I and other physicians understand the need to tailor the drug delivery vehicle to the active agent, to provide delivery of the active agent to where it is needed.

30. The instant application claims the oral administration, and colonic delivery, of agents which inactivate antibiotics. There are several reasons why it is important to administer the agents specifically to the colon. First and foremost, the development of antibacterial resistance in the commensal flora occurs mostly in the colon, because this is the segment of the intestine where almost all intestinal bacteria reside. The upper part of the intestine is populated with only very few bacteria. Most of antibiotics given orally are not fully absorbed through the gastrointestinal tract and there is a significant amount of "residual antibiotic" that reaches the colon. When residual antibiotics reach the colon, they will create a selective pressure that will select resistant bacteria among those composing the colonic commensally flora. They will also impact significantly on the protective commensal flora, inducing adverse effect such as diarrhea, pain etc. Many antibiotics are delivered orally, and are intended for systemic administration to treat systemic infection. Orally-administered antibiotics are absorbed throughout the gastrointestinal tract but mostly through the upper part of the tract, i.e. jejunum and ileum. If one were to deliver agents which inactivate the antibiotics too early (i.e., before the colon), the agents would interfere with the antibiotic therapy. However, by delivering the enzymes specifically to the colon, the present invention enables one to minimize the effect of the residual antibiotics on the development of antibacterial resistance and secondary adverse effects on commensal flora. Minimizing the effect of antibiotics on the

development of antibacterial resistance has been a focus of my research for a number of years, and it was not a trivial exercise to get to the point where we have developed a formulation to accomplish this goal.

31. It is worth noting that antibiotics can also be administered via different routes than the oral route. For example, intravenous antibiotics go right into the blood stream, and are thus delivered systemically without passing through the stomach. However most antibiotics that circulate in the blood are excreted at least in part through the liver and reach the intestinal tract through the biliary canal under active form. So, for intraveneously-administered antibiotics, one could think that it would not be necessary to deliver the agents specifically to the colon but that delivery in any part of the colon downstream the biliary canal would be adequate. However, the pH of the stomach and presence of many digestive enzymes in the jejunum and in the ileon would inactivate for sure, the delivered agent. Thus, the delivered agent would not be available anymore to inactivate the antibiotic in the colon. Enzymatic agents capable of inactivating antibiotics could, in theory, be administered anywhere after the stomach (where they would be degraded), and eventually work their way down to the colon. Since the antibiotic does not pass through the digestive tract, such non-specific delivery would not adversely affect the antibiotic therapy.

32. Thus, to understand the need for inactivating an antibiotic with an isolated active agent, one would a) need to appreciate the limitations associated with delivering a whole bacteria that expresses the active agent, and decide to instead administer an isolated agent, and b) understand how the antibiotic itself is administered, in order to determine an appropriate drug delivery vehicle to deliver the isolated active agent. The cited references do not provide any such understandings - these understandings were first provided by the present application.

33. Based on the facts outlined in this Declaration, the specification provides a sufficient description of "isolated" active agents to enable one to produce or purchase suitable active agents. The Olshenitsky reference cited in the novelty rejection fails to

disclose an isolated enzyme, and there is no evidence that the specific bacterial strain even produced such an enzyme. The secondary references are thus insufficient to overcome the faulty logic used in the novelty rejection. The references cited in the obviousness rejection do not provide any logical basis for placing an agent capable of inactivating an antibiotic into a colonic drug delivery vehicle, or even any reason to deliver such an agent to a patient. Even if there was a reason to administer the agent to a patient, there are a wide variety of drug delivery vehicles that could have been used, and the manner in which the antibiotic itself is administered is relevant to how the active agent must be administered. None of these factors is discussed in any of the cited references.

34. Finally, I note that the Examiner states that it would be obvious to administer isolated enzymes to a patient, because they are purportedly indigenous to the colon. The logical reasoning appears to be that while the cited references fail to teach *any* reason to administer the isolated enzymes to the colon, there would be motivation to orally administer anything present in the colon. Applying this logical reasoning, it would be just as obvious to orally administer fecal matter itself, because fecal matter is indigenous to the colon, and the bacteria present in the fecal matter would inherently produce the enzyme without requiring a further isolation process. This logical reasoning is flawed, because these enzymes are not indigenous to every patient. The genetic code for producing them is only found in bacteria present in a minority of patients who have, unfortunately, developed antibacterial resistance, and the enzymes are only produced in response to the presence of the antibacterial agent in the colon. Fortunately, not all patients have developed antibacterial resistance, and the present invention is directed to compositions and methods for preventing the development of such antibacterial resistance.

35. As a below-named Declarant, I hereby declare that all statements made herein of my own knowledge are true and that all statements made on information and belief are believed to be true; and further that these statements are made with the knowledge that willful false statements, and the like, so made are punishable by fine or imprisonment, or both, under

Section 1001 or Title 18 of the United States Code and that such willful false statements may jeopardize the validity of the application or any patent issued thereon.

Respectfully submitted,

Antoine Andrémont, M.D., Ph.D.

Dated: August 20, 2008